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Seroepidemiology of Human Parvovirus B19 in Taiwan

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In order to determine the prevalence and risk factors of human parvovirus B19 (B19) infection in Taiwan, a seroepidemiological study was carried out in 19 townships. Serum samples were collected from 862 healthy residents, who were selected by stratified random sampling from various study areas. They were chosen from four different ethnic groups including aborigines, Fukien Taiwanese, Hakka Taiwanese, and mainland Chinese. Serum samples were screened for B19 IgG antibody by indirect antibody capture enzyme-linked immunosorbent assay (ELISA) and B19 IgM by IgM antibody capture (MAC)-ELISA, respectively. The overall prevalence of anti-B19 IgG and anti-B19 IgM was 32.8% and 0.35%, respectively. The anti-B19 seropositive rate in females was significantly higher than that of males (36.4% vs. 29.4%, $P < .001$). The age-sex-adjusted seropositive rate in urban townships (39.9%) was higher than that in aboriginal townships (30.5%, $P < .001$). The seropositive rate increased significantly with age showing a dose–response relationship ($P = 0.0001$ based on a trend test). Blood transfusion was found to be associated with an increased seropositive rate showing a multivariate-adjusted odds ratios of 1.6. ***J. Med. Virol. 57: 169–173, 1999.***

KEY WORDS: human parvovirus B19; ELISA, capture ELISA; seroepidemiology

INTRODUCTION

Human parvovirus B19 (B19) belongs to genus *Erythrovirus* of the *Parvoviridae,* a family of small DNA viruses with single-stranded, linear genomes approximately 5 kb in length [Siegl et al., 1985]. B19 antigen was first discovered in the serum of healthy blood donors [Cossart et al., 1975]. The virus has a remarkable tissue-tropism for erythroid progenitor cells in human bone marrow [Brown et al., 1993]. Hence, the virus is associated with a wide range of clinical manifestations such as erythema infectiosum (EI) in children [Anderson et al., 1983], acute arthritis in adults [Reid et al., 1985; White et al., 1985], and aplastic crisis in patients with chronic hemolytic anemia [Serjeant et al., 1981; Rao et al., 1983; Shade et al., 1986]. B19 may also cause chronic infection in immunocompromised patients leading to persistent anemia [Pattison et al., 1981]. The virus has also been implicated as a cause of fetal hydrops and fetal loss during pregnancy [Brown et al., 1984]. Moreover, B19 may spread through the transfusion of contaminated blood products [Williams et al., 1990].

Serosurveys for B19 antibodies have been reported for Fukuoka, Japan [Nunoue et al., 1985], U.S.A. [Anderson et al., 1986], West Germany [Schwarz et al., 1987], England and Wales [Cohen and Buckley, 1988], and Hong Kong [Lim et al., 1997]. Antibody prevalence ranged from 20% to 80%, depending on the simultaneous occurrence of B19 outbreaks. The prevalence of B19 virus infection and its associated risk factors in Taiwan have never been reported. To estimate the prevalence of B19 virus infection in Taiwan, serum samples collected from 862 healthy subjects recruited in 1989 were tested for the presence of B19 virus IgG and IgM antibodies.

MATERIALS AND METHODS

Study Townships and Subjects

The selection of study townships and subjects have been described in detail previously [Wang et al., 1988]. In brief, the general population in Taiwan was first stratified according to ethnic characteristics (i.e., aborigines, Fukien Taiwanese, Hakka Taiwanese, and

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mainland Chinese) and residential areas (i.e., northern, southern, eastern and western islands, and offshore islets). Because there were no Hakka Taiwanese living in the offshore islets, there were only 19 strata in this study (Ministry of the Interior of R.O.C., 1985). One residential township was randomly chosen from each stratum. Study subjects were selected from the name lists updated by household registration offices using a multistage random sampling method [Wang et al., 1988]. A subset sample of 862 study subjects were selected from the original samples of 7,278 for the examination of B19 seromarkers.

Serum Collection and Questionnaire Interview

The serum samples and information regarding risk factors were collected by public health nurses in local health centers in study townships. A 10-ml blood sample was collected from each study subject and serum samples were separated and stored at −70°C until examinations. A standardized interview with a structured questionnaire was used to obtain information on risk factors including sociodemographic characteristics and disease history.

B19 IgG Antibody Detection

The B19 IgG antibody was detected by indirect antibody capture enzyme-linked immunosorbent assay (ELISA) according to the manufacturer's instructions (DENKA SEIKEN, Tokyo, Japan). Briefly, 100 μl each of standards and 1:201 diluted specimens were added to viral antigen-coated microtiter plate wells. After incubation at room temperature for 1 hr, the wells were washed and 100 μl peroxidase-labeled goat antibody against human IgG was added and incubated for another 1 hr at room temperature. After washing, 100 μl substrate solution (3,3′,5,5′-tetramethyl benzidine) was added and the color was developed for 30 min at room temperature. After adding 100 μl stop reagent (0.6 N sulfuric acid) the optical density was read within 30 min at the dual filter wave length of 450/630 nm with an automatic plate reader. Cut-off values for a positive test were calculated as described by the manufacturer.

B19 IgM Antibody Detection

The B19 IgM antibody was detected by MAC-ELISA according to the protocols of the manufacturer (DENKA SEIKEN, Tokyo, Japan). Briefly, 100 μl each of standards and 1:201 diluted specimens were added to antihuman IgM antibody-coated microtiter plate wells. After a 1-hr incubation at room temperature, the wells were washed and 100 μl viral antigen was added and incubated at room temperature for 1 hr. After washing, 100 μl peroxidase-labeled goat antibody against human B19 was added and incubated for another 1 hr at room temperature. The successive procedures were the same as those in B19 IgG antibody detection.

Statistical Analysis

Odds ratios (ORs) and 95% confidence intervals (CIs) were calculated by unconditional logistic regression to examine the association between B19 infection and potential risk factors after adjustment for potential confounding factors including sex, age, and residential area [Breslow and Day, 1992]. The P value for trend was calculated as level of significance of regression coefficient (indicative of amount of increase in risk per unit change in prevalence) for each ordinal variable in logistic regression model with relevant adjusting variables.

RESULTS

Seropositive Rate of B19 Antibody by Sociodemographic Characteristics

Two hundred eighty-one of the 862 study subjects were seropositive for B19 IgG antibody and negative for IgM; three male subjects were seropositive for B19 IgM, (one of whom was negative for IgG and positive for IgM and two of whom were positive for both IgG and IgM). The seropositive rates of both IgG and IgM for males and females were 29.6% (130/439) and 36.4% (154/423), respectively (Table I). Thus the total positive rate was 32.9%. The overall B19 seropositive rates in aboriginal, rural, and urban townships were 30.5%, 28.5%, and 39.9%, respectively. In all residential areas, females had a higher seropositive rate than males, and both males and females in urban townships had the highest seropositive rates. The seropositive rate in females remained significantly higher than that of males ($P < .05$) after adjustment for age and residential area. The seropositive rates were found to increase significantly with age and ranged from 10.3% in ⩽14-year-old group to 51.3% in ⩾60-year-old group. The older the subject, the higher the seropositive rate after adjustment for sex and residential area ($P < .001$). Residents in urban townships had a significantly higher seropositive rate than those in aboriginal townships ($P < .05$) after adjustment for age and sex. All study subjects were classified into three groups according to their educational levels, that is, senior high or above (203 subjects), junior high (169 subjects), and elementary and illiteracy (490 subjects) groups. The results showed that there was no significant difference in seropositive rate at different educational levels.

B19 Antibody Seropositive Rates by Blood Transfusion and Tattooing

As shown in Table II, those subjects with blood transfusion histories had a higher seropositive rate (53.6%) than those without (31.9%), after adjustment for age, sex, and residential area. There was no significant association between seropositivity of B19 and tattooing history.

Multiple Regression Analysis of Risk Factors Associated With Seropositivity of B19 Antibody

In the multiple logistic regression analysis, age, sex, residential area, and blood transfusion history were included in the regression model. As shown in Table III, age, sex, and residential area remained significantly associated with seropositivity of B19 antibody.

TABLE I. Associations With Seropositivity of Antibody Against Human Parvovirus B19 for Demographic Characteristics in Taiwan, 1989

Variable	Group	No. tested	No. positive (%)	OR (95% CI)	Adjusted OR (95% CI)
Sex	Male	439	130 (29.6)[d]	1.0 (referent)	1.0[a] (referent)
	Female	423	154 (36.4)	1.4* (1.0–1.8)	1.4* (1.0–1.9)
Age (years)	⩽14	116	12 (10.3)	1.0 (referent)	1.0[b] (referent)
	15–19	126	20 (15.9)	1.6 (0.8–3.5)	1.6 (0.8–3.5)
	20–29	115	27 (23.5)	2.7** (1.3–5.6)	2.6* (1.3–5.5)
	30–39	136	49 (36.0)	4.9*** (2.4–9.8)	4.8*** (2.4–9.7)
	40–49	132	63 (47.7)	7.9*** (4.0–15.7)	7.8*** (3.9–15.5)
	50–59	120	53 (44.2)	6.9*** (3.4–13.8)	6.6*** (3.3–13.3)
	⩾60	117	60 (51.3)	9.1*** (4.5–18.4)	9.1*** (4.5–18.5)
	Trend test			$P < .001$	$P < .001$
Residential township	Aboriginal	295	90 (30.5)	1.0 (referent)	1.0[c] (referent)
	Rural	284	81 (28.5)	0.9 (0.6–1.3)	0.9 (0.6–1.3)
	Urban	283	113 (39.9)	1.5* (1.1–2.1)	1.5* (1.1–2.2)

OR, odds ratio; CI, confidence interval.
[a]Adjusted variables included: age, residential township.
[b]Adjusted variables included: sex, residential township.
[c]Adjusted variables included: sex, age.
[d]Include either IgG positive or IgM positive cases.
*$P < .05$; **$P < .01$; ***$P < .001$.

TABLE II. Associations With Seropositivity of Antibody Against Human Parvovirus B19 for History of Blood Transfusion and Tattooing in Taiwan, 1989

Variable (95% CI)	Group	No. tested	No. positive (%)	OR (95% CI)	Adjusted OR[a]
Blood transfusion	No	808	258 (31.9)	1.0 (referent)	1.0[a] (referent)
	Yes	54	26 (53.6)	2.0* (1.1–3.4)	1.7** (1.0–3.1)
Tattoo	No	810	264 (32.6)	1.0 (referent)	1.0 (referent)
	Yes	52	20 (38.5)	1.3 (0.7–2.3)	1.1 (0.6–2.1)

OR, odds ratio; CI, confidence interval.
[a]Adjusted variables included: sex, age, and residential status.
*$P < .05$; **$P < .10$.

TABLE III. Multivariate-Adjusted Odds Ratios for Various Risk Factors Associated With Seropositivity of Antibody Against Human Parvovirus B19 in Taiwan, 1989

Variable	Group	Multivariate-adjusted odds ratio (95% CI)
Sex	Male	1.0 (referent)
	Female	1.4* (1.0–1.9)
Age (yr)	⩽14	1.0 (referent)
	15–19	1.6 (0.7–3.5)
	20–29	2.6* (1.2–5.5)
	30–39	4.7** (2.3–9.4)
	40–49	7.4** (3.7–14.8)
	50–59	6.4** (3.3–13.5)
	⩾60	8.9** (4.4–18.0)
Residential status	Aboriginal townships	1.0 (referent)
	Rural townships	0.9 (0.6–1.4)
	Urban townships	1.5* (1.1–2.2)
Blood transfusion history	No	1.0 (referent)
	Yes	1.6*** (0.9–2.8)

CI, confidence interval.
*$P < .05$; **$P < .01$; ***$P < .10$.

The multivariate-adjusted OR was 1.4 for females compared with males, whereas residents in urban townships had a 1.5-fold seropositivity rate compared with residents in aboriginal townships. The older the subject, the greater the multivariate-adjusted OR, which was as high as 8.9 for those older than 60 years as compared with those younger than 15 years. The multivariate-adjusted OR was 1.6 for blood transfusion history.

DISCUSSION

B19 was first identified and characterized from serum samples that were assayed for hepatitis B virus. The virus was then reported to be associated with the occurrence of aplastic crisis in patients with chronic hemolytic anemia, fifth disease, still births [Knott et al., 1984], arthritis, and persistent infection [Kurtzman et al., 1989; Anderson, 1990; Brown et al., 1994; Pattison, 1994; Musiani et al., 1995; von Poblotzki et al., 1995; Cassinotti et al., 1997; Gray et al., 1998]. It has also been associated with the contamination of blood products [Erdman et al., 1997].

B19 infection has been reported in many countries around the world. The seropositive rate of B19 IgG antibody varies by location and time of the last B19 epidemic. In Japan, the seropositive rate was reported to be 20% in the young age groups of <20 years and >80% in the old age groups of >60 years [Yamashita et al., 1992]. In England and Wales, the seropositive rate was 5–15% in the young age groups of 1–5 years, 50–60% in older children, young adults, and women of childbearing age, and >85% in the old age group of >70

years. In the US, the seropositive rate was 2% in young age group of <5 years and 49% in adults aged >20 years.

In Taiwan, a small outbreak of EI was observed last year (personal communication), but before 1989 no outbreaks of B19 infection had been reported. The seropositive rate of B19 antibody was found to be 10.3% in young age group of ⩽14 years and 51.3% in old age group of ⩾60 years. The increasing seropositive rate with age is consistent with those reported in other countries [Cossart et al., 1975; Paver and Clarke, 1976; Edwards et al., 1981; Mortimer et al., 1983; Courouce et al., 1984; Okabe et al., 1984; Anderson et al., 1986; Cohen and Buckley, 1988; Yamashita et al., 1992; Lim et al., 1997].

In this study lower prevalence rate (23.5%) was found in young adults (20–29 years). This result is similar to those observed in Japan: <20% in the 20–29-year-old group [Yamashita et al., 1992] and Hong Kong (9.5% in the 15–24-year-old group [Lim et al., 1997]), but different from those in the western countries with high prevalence rate of 49% in US [Anderson et al., 1986] and 50–60% in the UK in the young age group [Cohen and Buckley, 1988]. These results suggest that only fewer sporadic cases or asymptomatic infection of B19 might have occurred in the Far East than western countries.

The higher prevalence rate in urban areas in this study may have been because of the crowdedness of the population in the urban areas, because human parvovirus B19 is transmitted effectively after close contact exposure. This result might be attributed to the possible mode of B19 transmission by aerosol via the respiratory tract. No significant difference in B19 infection rates was observed at different educational levels.

It has been reported that B19 infection can be transmitted through parenteral routes [Musiani et al., 1995]. In this study, blood transfusion history was associated with an increased seropositive rate of B19, but no association was observed between B19 infection and tattooing history.

It is interesting that a significant difference was found in this study with a higher positive rate among females (<0.05), although no gender difference was found in Japan [Matsunaga et al., 1995]. A further study is necessary to elucidate this difference.

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